NON-TECHNICAL ABSTRACT OF PROTOCOL

Malignant gliomas recurring after radiation and/or chemotherapy are uniformly fatal, and current approaches to treatment only briefly prolong survival. For these patients, a new approach to treat their cancer (tumor) will be tested. In this approach, cells taken from normal skin (fibroblasts) are engineered to produce a chemical stimulant for the immune system (cytokine), named IL-4. A virus carrying the gene for IL-4 is used to transfect the fibroblasts, leading to production of IL-4 by the infected fibroblasts. These IL-4 producing fibroblasts are mixed with a vaccine preparation consisting of tumor cells taken from the patient and grown in cell culture, and the most powerful immune stimulating cells, dendritic cells. Animal studies have shown that a vaccine made of dendritic cells (DC) and a glioma cell line, which is genetically identical to the dendritic cells in expression of immunologically important molecules(syngeneic) will stimulate host immunity against the tumor from which the cultured tumor cells were derived. In this proposed study, the IL-4 expressing fibroblasts are added to the vaccine preparation, because animal studies have shown that this stegnthens the immune response over that produced by dendritic cells and tumor cells mixed together by themselves. When DC were co-cultured with UV-irradiated glioma cells and used to innoculate rats with intracranial gliomas intradermally in a flank along with IL-4 fibroblasts, the animals treated with the combined vaccine survived longer than the animals who did not receive the IL-4 producing fibroblasts as part of the vaccine. Studies in animals suggest several reasons for the improved performance of the vaccine which includes dendritic cells and IL-4 producing fibroblasts:

1. Injecting DC vaccine under the skin has been shown to increase the responses of the animal's T cell response that kills glioma cells in a specific manner.
2. Addition of fibroblasts secreting IL-4 appears to enhance the immuno-reactivity against the glioma cells, at least partially by increasing the number of glioma-antigen loaded DCs that migrate to the lymphoid organs.

This protocol is designed to test the vaccine protocol which has been effective in experimental animals in patients.

Patients with malignant recurrent gliomas after standard treatment will have a piece of their brain tumor surgically removed, during an operation performed as part of the standard clincial care of the patient, and brought to the laboratory. In the laboratory, the brain tumor cells will be irradiated with ultra violet (UV) in order to induce apoptosis, a form of cell death. The patients will also undergo leukapheresis via peripheral vein to collect lymphocytes that will serve as source of DC. Leukophoresis is a procedure which resembles kidney dialysis, in which the white cells are taken from the patients blood, and the rest of the blood cells and plasma is returned automatically to the patient. DCs will be cultured form these cells and co-cultured overnight with UV-irradiated glioma cells. During the same operation at which the brain tumor is removed, a piece of skin will be also harvested from the patients' abdomen and will be dissociated to make single cell suspension in the laboratory. These cells will be cultured and will be transfected with the retroviral vector encoding IL-4. The vaccine will consist of DC co-cultured with UV-irradiated apoptotic glioma cells and IL-4 transfected fibroblasts. Fibroblasts will be admixed with DC immediately before injection to the patients. Patients will be innoculated with the vaccine 2 different times, 2 weeks apart. The effectiveness of the vaccine will be studied in several ways. A week before the first vaccine the patients will be injected with irradiated tumor cells intradermally in order to observe inflammation at the injection sites. Then a similar injection of tumor cells will be performed a week after the second vaccination step. The difference in inflammation and induration (swelling) between the prevaccination and post-vaccination tumor cell injections is a reflection of the stregnth of the immune response stimulated by the vaccinations. Increased induration after vaccination would indicate induction of cellular immune response. White blood cells are taken from peripheral blood and will be analyzed for the presence of immune cells reacting against tumor cells. We will also look for immune cells reacting to the tumor cells at the vaccination sites in the thigh. These specific antitumor immune cells will be stimulated to devide in cell culture, using a previously optimized growth stimulating process involving another cytokine, IL-2, so that large numbers

of immune cells capable of killing tumor cells may be obtained. The patients are examined and will undergo brain imaging with MRI scanning at close intervals after vaccination to determine if there is improvement in the symptoms related to the brain tumor, and to monitor for side effects of the vaccine.